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Profiles of C- and N-trace gas production in N-saturated forest soils

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Abstract

This study provides for the first time data on the stratification of NO and N_2O production with soil depth under aerobic and anaerobic incubation conditions for different temperate forest sites in Germany (spruce, beech, clear-cut) and the Netherlands (Douglas fir). Results show that the NO and N_2O production activity is highest in the forest floor and decreases exponentially with increasing soil depth. Under anaerobic incubation conditions NO and N_2O production was in all soil layers up to 2–3 orders of magnitude higher then under aerobic incubation conditions. Furthermore, significant differences between sites could be demonstrated with respect to the magnitude or predominance of NO and N_2O production. These were driven by stand properties (beech or spruce) or management (clear-cut versus control). With regard to CH_4 the most striking result was the lack of CH_4 uptake activity in soil samples taken from the Dutch Douglas fir site at Speulderbos, which is most likely a consequence of chronically high rates of atmospheric N deposition. In addition, we could also demonstrate that CH_4 fluxes at the soil surface are obviously the result of simultaneously occurring uptake and production processes, since even under aerobic conditions a net production of CH_4 in forest floor samples was found. The provided dataset will be very useful for the development and testing of process oriented models, since for the first time activity data stratified for several soil layers for N_2O, NO, and CH_4 production/oxidation activity for forest soils are provided.

1. Introduction

Soils are of significant importance as sources or sinks of environmental important atmospheric trace gases such as CH_4, N_2O and NO (e.g. IPCC, 1997; Davidson and Kingerlee, 1997; Smith et al., 2000). The exchange rates of these trace gases between soils and the atmosphere are mainly driven by a few microbiological processes in the soil. With regard to NO and N_2O nitrification and denitrification are the key mi-

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crobial processes responsible for production, but also consumption of these both trace gases in soils (Conrad, 2002; Robertson and Tiedje, 1987), whereas methanogenesis and CH_4-oxidation are the two processes controlling CH_4 soil-atmosphere fluxes (Conrad, 1996). The magnitude of N and C trace gas exchange and the direction of flux, i.e. deposition or emission, is strongly controlled by the aeration status of the soil. Under anaerobic conditions soils can act e.g. as a source for atmospheric CH_4, whereas under aerobic conditions most soils tend to be a sink for atmospheric CH_4 (Butterbach-Bahl and Papen, 2002). Though the exchange of N_2O and NO at the soil surface is also the result of simultaneously occurring production and consumption processes in the soils, soils have predominantly been found to be net emission sources for N_2O and NO (e.g. Bremner, 1997; Davidson and Kingerlee, 1997). Changes in the aeration status directly affect the contribution of nitrification and denitrification to the observed net-N trace gas emissions (Bollmann and Conrad, 1998). Under aerobic conditions nitrification may contribute significantly to N_2O and especially NO production, whereas under prevailing anaerobic conditions denitrification is the main process of N trace gas production (Bateman and Baggs, 2005). Several field and laboratory studies have shown that under non-substrate limiting conditions N trace gas production under anaerobic conditions tend to be up to several orders of magnitude higher than under aerobic soil conditions (Bollmann and Conrad, 1998). However, there is still only limited information available to which extend the aeration status will change C and N trace gas exchange under controlled environmental conditions. Furthermore, most of such studies have been performed for agricultural or grassland soils rather than for forest soils.

This study was intended to complement field measurements of N and C trace gas exchange at two different forest locations in the Netherlands and Germany, which were carried out in the framework of the EU-funded NOFRETETE project. Both forest sites, i.e. Höglwald in South Germany and Speulderbos in the Netherlands, are exposed to high loads of atmospheric N deposition (20–50 kg N ha^{-1} yr^{-1}) and have been shown previously to be strong emitters of NO (Gasche and Papen, 1999; van Dijk and Duyzer,

1999) and, with regard to Höglwald, also of N_2O (Butterbach-Bahl et al., 2002a). Furthermore, soils at Höglwald were also found to be significant net sinks for atmospheric CH_4 (Butterbach-Bahl and Papen, 2002). In the context of this study we aimed to characterize the vertical distribution of C and N trace gas production and consumption at both locations and to identify how atmospheric N deposition, forest type (beech, spruce, Douglas fir) or forest management (clear-cutting) may have affected the net exchange of NO, N_2O and CH_4.

2. Materials and methods

2.1. Study sites

Stratification of N_2O, NO and CH_4-trace gas production and consumption in forest soils was done for four different forest sites in Germany and the Netherlands. Three of the four sites were located at Höglwald, Germany, where stands of mature beech and mature spruce as well as a recently cleared spruce stand (clear cut in March 2000) were investigated. The latter site is in the following referred to as clearcut site. The area of Höglwald is located in South Germany approx. 40 km west of the city of Munich and is characterized by high atmospheric N deposition in the range of 15 (clear cut) to 30 kg N ha^{-1} a^{-1} (Table 1). The fourth site was located at Speulderbos, Netherlands. The investigated Douglas fir plantation receives approx. twofold higher amounts of atmospheric N deposition than compared to Höglwald (Table 1). Further information on main climatic and soil characteristics is provided in Table 1.

2.2. Determination of rates of microbial NO, N_2O and CH_4 production/consumption

In order to determine changes in rates of microbial N and C trace gas production and consumption with soil depth, stratified soil samples were taken at all sites. The spruce and clearcut sites at Höglwald were sampled in November 2000, April 2001, August

2002 and January 2004. The Douglas fir site at Speulderbos and the beech site at Höglwald were sampled in October 2003 and January 2004, respectively. An overview about the stratification of soil sampling at the different sites is provided in Table 1. With the exception of the beech site at Höglwald, where the forest floor is <0.02 m, the forest floor was subdivided into two sampling horizons (Table 1). All soil samples were transferred to the microbiological laboratory of IMK-IFU at Garmisch-Partenkirchen where analysis was started within a few days after sampling. Soil samples were pre-incubated at 10°C two days prior of the experiments. All experiments were carried out at 10°C. For all sampling dates soil moisture values were highest for forest floor samples (>100% w/w basis) and lower for soil samples taken from the mineral soil (in average 40–60% w/w soil moisture for samples taken from 0–0.1 m soil depth and 15–30% w/w soil moisture for samples taken from 0.1–0.35 m soil depth).

Soil samples of a given horizon were further subdivided into 3 subsamples. For subsamples taken from the forest floor 20 g, for subsamples taken from the mineral soil horizon from 0–0.05 m soil depth 50 g and for all other soil depths 100 g of soil were filled into 350 ml glass flasks. For aerobic incubations these flasks were closed with butyl rubber stoppers, and the change in concentrations of N_2O, NO and CH_4 in the headspace of the flasks was followed for the next 1–2 h in intervals of 15–20 min for NO, and 6–8 h in hourly intervals for N_2O and CH_4, respectively, by taking 3 ml air samples from the headspace of the flasks with gas tight syringes. Air samples were immediately analysed for the respective concentrations of the investigated trace gases (see below). The same procedure was also followed for anaerobic incubations. However, for these experiments the headspace air was exchanged versus a pure N_2 atmosphere immediately after closure of the flasks as described by Butterbach-Bahl et al. (1997) and Butterbach-Bahl and Papen (2002). Production or consumption rates were calculated on a soil dry weight basis (SDW) from the linear increase of N and C trace gas concentrations with time (see Butterbach-Bahl and Papen, 2002).

Concentrations of CH_4 and N_2O in air samples were determined by gas chromatography using either a flame ionization detector (FID) for the detection of CH_4 or a ^{63}Ni

electron capture detector for the detection of N_2O. Analytical conditions are described in detail by Papen and Butterbach-Bahl (1999) and Butterbach-Bahl and Papen (2002). NO concentrations in air samples were detected by injecting 3 ml of gas sample in the sampling gas flow of a chemoluminescence detector (CLD 770 AL ppt, Ecophysics AG, Dürnten, Switzerland). As sampling gas NO-free synthetic air (Messer Griesheim, Olching, Germany) at a flow rate of approx. 500 ml min^{-1} was used. All instruments were routinely calibrated either hourly (N_2O and CH_4) or every other day (NO) with calibration standards (Messer Griesheim, Olching, Germany).

2.3. Auxiliary measurements

The pH of soil samples was measured after addition of 50 ml of aqueous 0.1 M (?) $CaCl_2$ solution to 10 g soil and vigorous shaking for 15 min. NH_4 and NO_3 concentrations in soil samples taken from the spruce and clearcut plots at Höglwald were determined by extracting soil subsamples (5 g) with 50 ml 0.1 N $KAl(SO_4)_2$. The suspension was shaken for 20 min, and centrifuged for 15 min at 4 °C and 12 000 g (Beckmann Instruments, München, Germany). The supernatant was filtered through 0.2 μm membrane filters (Millipore, Frankfurt) and analyzed immediately for NH_4^+ and NO_3^- by ion chromatography (DIONEX DX 500; anion-column: AS4A; cation column: CS12; DIONEX Company, Idstein, Germany). For the determination of microbial biomass C the fumigation extraction method as described by Vance et al. (1987) was used. All experiments were performed at least in triplicate.

2.4. Statistical analyses

All statistical analyses were performed with SPSS 8.0 (SPSS Inc., US) and Microcal Origin 6.1. Tests of significance of differences between specific rates of C and N trace gas production were performed by using the multiple range test (LSD) by ANOVA.

3. Results

3.1. Vertical distribution of microbial biomass, soil pH and inorganic N

At all sites soil pH values were well below 5.0 and showed a typical check mark shape, with most acidic conditions in the uppermost mineral layer, and significantly or slightly higher pH values in the forest floor and in deeper soil layers of the mineral soil (Fig. 1). Most acidic conditions were observed for the Speulderbos site, where for the uppermost mineral soil a pH of only 2.7 was found. On the other hand, at this site pH values increased most significantly with soil depth to values well above 4.1 at a soil depth of 0.35 m. For the Höglwald sites, soil pH values were lowest for the spruce site and the recently clear-felled spruce site. Soil pH values of the beech site were for all soil layers at least half a pH value higher than compared to respective soil layers of the spruce and clear-felled sites, respectively. Such pronounced differences in soil pH between spruce and beech stands at Höglwald, which are mainly due to differences in cation cycling and atmospheric N input have already been reported previously (Rothe et al., 2002).

Ammonium and nitrate concentrations in different soil depths were only measured for the spruce and the clear-felled sites at Höglwald. Figure 2 shows that in average over all sampling dates ammonium concentrations were highest in the forest floor with values >7 μg N g^{-1} SDW and decreased exponentially to values <0.7 μg N g^{-1} SDW with increasing soil depth. No significant differences were found in ammonium concentrations between sites and a given soil layer. Also with regard to nitrate concentrations in soil samples, highest values were found in the forest floor (>10 μ g N g^{-1} SDW). Somewhat lower concentrations of nitrate were found in the mineral soil, but in contrast to ammonium concentrations the decrease in nitrate concentrations with soil depth was less pronounced (e.g. 30 cm soil depth: 4 μg nitrate N g^{-1} SDW) (Fig. 2). Compared to the spruce control site nitrate concentrations at the clear-felled site were significantly lower for forest floor samples (p<0.05). For all other soil layers no significant differences were found

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At all sites investigated the amount of microbial biomass C decreased exponentially with soil depth from up to 10 mg C g^{-1} SDW in the forest floor to <0.02 mg C g^{-1} SDW in 0.3–0.4 m soil depth (Fig. 3). For all soil depths microbial biomass C values obtained for the beech site at Höglwald were at least a factor of two higher than compared to the other sites ($p<0.05$). Microbial biomass was not significantly different between the spruce and clear-felled sites at Höglwald and the Douglas fire site at Speulderbos. However, of all sites investigated the Speulderbos site tended to show lowest values of microbial biomass C in the mineral soil (Fig. 3).

3.2. Vertical distribution of N_2O and NO production in the soil profiles

At all sites N_2O production was found to be highest in the forest floor (Fig. 4). Mean N_2O production in the forest floor of the Höglwald beech site was 28.3±9.7 ng N_2O-N g^{-1} SDW h^{-1}, whereas N_2O production in the forest floor of the other sites was significantly lower (Höglwald, spruce: 0.6–3.3 ng N_2O-N g^{-1} SDW h^{-1}, Höglwald, clearcut: 0.4–0.8 ng N_2O-N g^{-1} SDW h^{-1}; Speulderbos, Douglas fir: 0.1–6.0 ng N_2O-N g^{-1} SDW h^{-1}). Except for the 0.25 m layer of the mineral soil at the Höglwald beech site N_2O production in the mineral soil layers was mostly one magnitude lower than compared to the forest floor with values <0.2 ng N_2O-N g^{-1} SDW h^{-1}. This vertical stratification of N_2O production was even more pronounced for the Douglas fir site at Speulderbos, where N_2O production in the mineral soil was approx. three orders of magnitude lower than in the forest floor (Fig. 4). Mean specific N_2O production, i.e. ng N_2O-N production per mg microbial biomass C, was highest for the Höglwald beech site with a mean value of 3.5±2.9 ng N microbial biomass C h^{-1} and lowest for the Speulderbos Douglas fir site (0.4±0.2 ng N microbial biomass C h^{-1}). However, site differences were not significant on a $p<0.05$ level (Table 2).

Under anaerobic incubation conditions N_2O production increased by at least 1–2 orders of magnitude than compared to aerobic incubation conditions. Also under anaerobic incubation conditions highest N_2O production was observed with values >100 ng

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N_2O-N g^{-1} SDW h^{-1} for soil samples taken from the forest floor (Fig. 5). In contrast to results obtained for aerobic incubation conditions anaerobic N_2O production in soil samples taken either from the forest floor or from the uppermost 0.2 m of the mineral soil of the Höglwald spruce or Höglwald clearcut site was significantly higher than compared to soil samples taken from the Höglwald beech or Speulderbos Douglas fir site. Except for the Höglwald beech site specific N_2O production under anaerobic incubation conditions at all other sites was approx. two orders of magnitude higher than under aerobic incubation conditions, whereas this difference was only a factor of two for the Höglwald beech site (see Table 2).

The vertical stratification of NO production with soil depth was comparable to the results obtained for N_2O production, i.e. aerobic NO production was highest in the forest floor and sharply decreased in soil samples taken from the mineral soil (Fig. 6). Compared to aerobic N_2O production aerobic NO production in the forest floor was significantly higher and reached up to 371 ng NO-N g^{-1} SDW h^{-1} (Höglwald spruce site). Specific NO production was with a value of 130.6±57.7 ng N microbial biomass C h^{-1} significantly higher at the Höglwald spruce site than compared to the other sites (3.3–32.4 ng N microbial biomass C h^{-1}) (Table 2). Except for the Höglwald beech site specific NO production was at least one magnitude higher than specific N_2O production, thus showing that NO is produced in larger quantities in the soil as N_2O.

As was also found for N_2O production anaerobic incubation of soil samples strongly enhanced NO production (Fig. 7). Peak values of anaerobic NO production as e.g. observed for the Höglwald spruce site were with 1494 ng NO-N g^{-1} SDW h^{-1} in the same range as anaerobic N_2O production. Anaerobic NO production in the mineral soil was at least one magnitude lower in the mineral soil and decreased to values <40 ng NO-N g^{-1} SDW h^{-1} for all soil samples taken from soil layers deeper than 0.02 m. Specific NO production under anaerobic incubation condition was significantly highest for the Höglwald spruce site (848 ng N mg^{-1} microbial biomass C h^{-1}) (Table 2), which is in accordance with the results for aerobic incubation conditions. Lowest specific NO production under anaerobic but also under aerobic incubation conditions were observed

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for the Höglwald beech site.

3.3. Vertical distribution of CH_4 production and consumption in the soil profiles

Under aerobic incubation conditions the direction of CH_4 flux, i.e. net-uptake or net-production, was found to depend on the soil depth from which samples were taken. In most cases soil samples taken from the forest floor, i.e. predominantly organic material, showed a small net CH_4 production (range: −0.015–0.546 ng CH_4-C g^{-1} SDW h^{-1}) (Fig. 8). In contrast, soil samples of the uppermost layers of the mineral soil were found to be a net sink for CH_4 at the Höglwald sites. Figure 8 shows that CH_4 uptake at the Höglwald beech site was with −0.62±0.01 ng CH_4-C g^{-1} SDW h^{-1} at least 5 times higher than CH_4 uptake rates in the mineral soil at the Höglwald spruce and clearcut sites, which are in direct vicinity of the Höglwald beech site (100 m distance). At all sites at Höglwald CH_4 uptake activity in the mineral soil decreased with increasing soil depth. However, under aerobic incubation conditions the soil was still a weak sink for CH_4 even at 0.35 m (Fig. 8). In contrast to the Höglwald sites there was no significant net uptake of CH_4 by the mineral soil of the Speulderbos Douglas fir site. If mineral soil samples were derived from soil layers >0.1 m even a small net production of CH_4 under aerobic incubation conditions could be observed. Therefore, the mean specific CH_4 exchange rate at the Speulderbos site was positive (net production across the soil profile) with 0.02±0.06 ng C mg^{-1} microbial biomass C h^{-1}, and not negative (net uptake across the soil profile) than for the Höglwald sites (−1.56 to −0.46 ng C mg^{-1} microbial biomass C h^{-1}) (Table 2).

Under anaerobic incubation conditions CH_4 production in forest floor soil samples was found to be strongly enhanced (Fig. 9). CH_4 production in the forest floor at the Höglwald sites was in a range of 0.5–1.5 ng CH_4-C g^{-1} SDW h^{-1}, whereas CH_4 production in the forest floor of the Speulderbos Douglas fir site was significantly lower with values <0.1 ng CH_4-C g^{-1} SDW h^{-1}. CH_4 production in mineral soil samples taken at soil depths <0.05 m were at least one order of magnitude lower than com-

pared to CH_4 production in the forest floor. As was found for CH_4 uptake under aerobic incubation conditions, also under anaerobic incubation conditions and for CH_4 production in 0.065 m and 0.15 m soil depth a significant difference could be found between the beech site on the one hand (0.01–0.2 ng CH_4-C g^{-1} SDW h^{-1}) and the spruce or clearcut sites on the other hand (0.03–0.08 ng CH_4-C g^{-1} SDW h^{-1}). Even though the stratification of CH_4 production at the Speulderbos Douglas fir site was comparable to those found for the Höglwald sites, CH_4 production under anaerobic incubation conditions was in all soil layers approx. one order of magnitude lower than compared to the respective soil layers at the Höglwald sites (Fig. 9). Site differences diminished for the Speulderbos Douglas fir site and Höglwald beech site if mean specific rates of CH_4 production are calculated. With 0.07 ng C mg^{-1} microbial biomass C h^{-1} (Höglwald beech site) and 0.12 ng C mg^{-1} microbial biomass C h^{-1} (Speulderbos Douglas Fir site) these values were not significantly different, but significantly lower than the values for the Höglwald spruce and clearcut sites (>0.69 ng C mg^{-1} microbial biomass C h^{-1}) (Table 2).

4. Discussion

The amount of microbial biomass found at the coniferous forest sites at Speulderbos or Höglwald in the forest floor and first 0.05 m of the mineral soil are with 0.1–10 mg C g^{-1} SDW (approx. 1–16 g microbial C m^{-2}) in a comparable range than observed also for other nitrogen deposition affected temperate coniferous forest ecosystems in central Europe such as the Solling area (e.g. spruce: 4–5 g Microbial C m^{-2}; Corré and Lamersdorf, 2004). Throughout the profile higher values for microbial biomass were found for the beech site at Höglwald, but such a difference between spruce (coniferous) and beech (deciduous) sites was also reported for the Solling (Corré et al., 2003; Corré and Lamersdorf, 2004) and for other sites in Austria (Zechmeister-Boltenstern et al., 2002). Also observed ammonium and nitrate concentrations in soils at Höglwald are within reported ranges (nitrate: 1–30 ng N g^{-1}; Ammonium: 2–200 ng N g^{-1}) (Priha et

al., 1999; Herman et al., 2002; Wang and Ineson, 2003; Aubert et al., 2005). However, it is worthwhile to mention that soil nitrate concentrations at the clear-cut site were at least for the forest floor layers significantly lower than compared to the control site, and, thus, further support the explanation that mainly increased rates in soil water fluxes are responsible for observed increases in nitrate leaching (>30 kg N ha^{-1} yr^{-1}) after clear-cutting (Weiss, Technical University of Munich, personnel communication).

At all sites N_2O and NO production under aerobic and anaerobic incubation conditions was highest in the forest floor. Our laboratory results are in good agreement with results from field experiments at Höglwald, where soil layers were subsequently removed and where also the forest floor was identified as the layer contributing most to the actual N_2O (70%) or NO (79%) emissions (Gasche and Papen, 1999; Papen and Butterbach-Bahl, 1999).

The magnitude of aerobic N_2O production in the forest floors of our study sites (0.1–28 ng N_2O-N g^{-1} SDW) were in most cases much higher as production rates of N_2O under aerobic conditions in agricultural soils (Bollmann and Conrad, 1998, who reported rates in a range of 0.05–0.2 ng N_2O-N g^{-1} SDW). However, with regard to mineral soil samples differences in the aerobic N_2O production between the investigated forest soils (range in this study: 0.001–0.2 ng N_2O-N g^{-1} SDW) and agricultural (Bollmann and Conrad, 1998), or other meadow or forest soils (0.02–0.4 ng N_2O-N g^{-1} SDW; Gödde and Conrad, 2000) diminish. The range of N_2O production observed within this study for forest floor samples is close to those observed by others in fertilized soil incubations (0.3–600 ng N_2O-N g^{-1} SDW) (Parton et al., 1988; Bateman and Baggs, 2005; Kinney et al., 2005). The observed significantly lower N_2O production potential of the Speulderbos Douglas fir site than compared to the Höglwald sites mirrors results from field measurements. Also here, Speulderbos showed low rates of N_2O emissions. This is remarkable, since one would assume that the observed high rates of N deposition (approx. 50 kg N ha^{-1} yr^{-1}) not only enhance NO emissions, as observed already earlier by Van Dijk and Duyzer (1999), but also N_2O emissions. Since for this site nitrate concentrations in the soil solution are with 74–97 mg l^{-1} (Duyzer,

unpublished) even higher than for the Höglwald spruce site (approx. 40 mg l^{-1}, Rothe et al., 2002), we only can speculate that the reason for low N_2O emissions must be due to hydrological properties of the site. The Speulderbos forest site is a very well developed Douglas fir stand with a very dense canopy (LAI>9), thus losing considerable amounts of precipitation by interception. Furthermore, a thick forest floor prevents or limits the penetration of throughfall into the mineral soil. All these factors result in relatively low soil moisture conditions in the mineral soil, which means that N_2O production by denitrification would not occur due to water limitations or, to be more precise, due to the sufficient aeration of the soil limiting denitrification (Conrad, 1996). This hypothesis is further supported by our finding that under anaerobic incubation conditions the differences between the Speulderbos Douglas fir site and the Höglwald beech site disappears. Though, it still remains if compared to the spruce and clear-cut site. This shows that the Speulderbos site has a high potential for elevated N_2O emissions if the environmental conditions and here especially the soil hydrology would change, e.g. due to selected cutting or clear-cutting. Our laboratory data also suggest that changes in the hydrological conditions due to clear-cutting has not affected the anaerobic N_2O production potential. Figure 5 shows that the stratification of anaerobic N_2O production at the clear-cut site, on which previously spruce was grown, was down to 0.2 m soil depth not significantly different from the spruce site. Only for deeper soil layers anaerobic N_2O production was higher at the clear-cut site than compared to the spruce site. This can be explained with higher values of nitrate leaching at the clear cut site, which supports higher denitrification activities also in deeper soil layers. Even though general soil properties and meteorological conditions are identical (Rothe et al., 2002) for the Höglwald Forest sites, anaerobic N_2O production as well as specific anaerobic N_2O production remained significantly lower at the beech site than compared to the spruce and clear-cut sites. This is remarkable, since soil moisture conditions especially in spring and winter time at the beech site are higher than compared to the spruce site (Butterbach-Bahl et al., 2002b). For that reason one would assume that the beech site supports a larger denitrifying population in the soil, which would finally support a

higher potential of N_2O production under anaerobic conditions. First results on counts of denitrifying bacteria in the soil of the beech and spruce sites would also support this hypothesis (Papen, personnel communication). But, obviously higher counts of denitrifying bacteria and higher average soil moisture values for the beech site have not resulted in a higher anaerobic N_2O production potential. The reason for that is most likely associated to differences in average nitrate availability throughout the year, which is lower at the beech site than compared to the spruce site. Rothe et al. (2002) found for the Höglwald Forest that nitrate leaching under beech is close to zero, but approx. 20 kg N ha^{-1} yr^{-1} at the spruce site. This difference is partly due to differences in the magnitude of atmospheric N deposition, which is approx. 10 kg higher at the spruce site. But, the lower nitrate concentrations in the soil under beech are most likely also due to differences in nitrate uptake by roots of beech and spruce. Geßler et al. (1998) found for the Höglwald beech and spruce sites, that beech roots do still show nitrate uptake activity under conditions of nitrogen saturation, whereas spruce roots did not show any nitrate uptake activity at all.

The observed increase of N_2O production under anaerobic incubation conditions by two to three orders of magnitude than compared to aerobic incubation conditions is in agreement with the results on the oxygen dependency of N_2O production in agricultural soils by Bollmann and Conrad (1998). However, their maximum N_2O production under anaerobic conditions was around 100 ng N_2O-N g^{-1} SDW, whereas in our study we found values of up to 2000 ng N_2O-N g^{-1} SDW in forest floor samples taken from the Höglwald clear-cut site.

NO production activity in soils is mostly attributed to nitrification rather than to denitrification (Skiba et al., 1997; Conrad, 2002). Only if oxygen is depleted below values of 0.1–0.5% denitrification may become the dominating source for NO production in soils (Bollmann and Conrad, 1998). For two agricultural soils in Germany Bollmann and Conrad (1998) reported rates of aerobic NO production in a range of 0.5–2 ng NO-N g^{-1} SDW and for anaerobic incubation conditions rates of NO production of 100–200 ng NO-N g^{-1} SDW. Even in a study were soils were taken from recently fertilized agricul-

tural fields, NO production remained ≤20 ng NO-N g^{-1} SDW (Tortoso and Hutchinson, 1990). Also in an other detailed study were the influence of soil properties on the turnover of nitric oxide was investigated (Gödde and Conrad, 2000) NO production in mineral soil samples taken either from agricultural, meadow or forest sites remained lower than 15 soils ng NO-N g^{-1}. Compared to these values rates of NO production under aerobic or anaerobic incubation conditions were much higher for soil samples taken from the forest floor (aerobic: 7–320 ng NO-N g^{-1} SDW; anaerobic: 100–1500 ng NO-N g^{-1} SDW), but in the same range for samples taken from the mineral soil (aerobic: 0.05–10 ng NO-N g^{-1} SDW; anaerobic: 1–130 ng NO-N g^{-1} SDW). This underlines that especially coniferous forest soils exposed to high loads of atmospheric N deposition will act as strong sources of nitric oxide. At least for the Höglwald sites it can be summarized that the high rates of NO production especially in the forest floor are matched by high nitrification activities (Gasche et al., 2002). The observed site differences in NO production activity, i.e. Höglwald spruce>Höglwald clear-cut>Höglwald beech≈Speulderbos fire, is in agreement with results of field measurements (Gasche and Papen, 1999; van Dijk and Duyzer, 1999; Gasche, unpublished) and demonstrate the usefulness of laboratory studies for evaluating site differences. The reason for such high NO production potentials and NO emissions especially from coniferous forest soils are most likely manifold: a) due to high rates of N deposition nitrogen is not limited, b) acidic soil reaction in the forest floor than compared to neutral pH values in agricultural soils can support higher NO production rates (e.g. Yamulki et al., 1997; Ormecci et al., 1999; Kesik et al., 2005[1]), c) the accumulation of "fresh" litter support high microbial C and N turnover rates, and d) modest soil moisture conditions in the forest floor support high nitrification activities and due to its favourable diffusivity also reduce the likeliness of NO consumption via denitrification and nitrification (Dunfield and Knowles, 1997).

Our incubation studies on CH_4 dynamics revealed principal differences in CH_4 up-

[1]Kesik, M., Blagodatsky, S., Papen, H., and Butterbach-Bahl, K.: Effect of pH, temperature and substrate on N_2O, NO, and CO_2 production by Alcaligenes faecalis, J. Appl. Microbiol., submitted, 2005.

take and production between the Höglwald sites and the Speulderbos Douglas fir site. For the latter site CH_4 fluxes were insignificant under aerobic incubation conditions, whereas under such incubation conditions samples from the forest floor were significant net CH_4 producers in contrast to mineral soil samples which showed a significant net CH_4 uptake. The high potential of the forest floor to act as a net CH_4-production source even under well aerated conditions has also been described previously for some other temperate forest sites (Saari et al., 1997; Yavitt et al., 1990) and is due to the high mineralisation activity in the forest floor obviously creating strictly anaerobic microsites in which CH_4 production can occur. At all Höglwald Forest sites, i.e. beech, spruce, and clear-cut, highest CH_4-oxidation activity was found in 0–0.15 m soil depth, where CH_4-oxidation rates of up to -0.6 ng CH_4 g^{-1} SDW h^{-1} (beech), −0.13 ng CH_4 g^{-1} SDW h^{-1} (spruce) and −0.06 ng CH_4 g^{-1} SDW h^{-1} (clear-cut) were observed. The magnitude of CH_4-oxidation activity found for the Höglwald sites are in excellent agreement with values reported in the literature for the 0–0.05 m mineral soil layer of a spruce forest site in the Netherlands (−0.44 ng CH_4-C g^{-1} SDW h^{-1}, Saari et al., 1997) which is also – like Höglwald – exposed to high loads of atmospheric N-deposition or with previous results for the beech and spruce sites of Höglwald (Butterbach-Bahl and Papen, 2002). However, compared to rates of CH_4-oxidation with low atmospheric N-input in Finland (−1.43 ng CH_4 g^{-1} SDW h^{-1}, Saari et al., 1997) or the Black Forest region in Germany (Steinkamp et al., 2001), the CH_4-oxidation activity in our soil samples was approx. 2–3 times lower. This finding, as well as the absence of CH_4 uptake activity at the Speulderbos site strongly support the hypothesis that chronic atmospheric N-deposition will strongly decrease the CH_4-oxidation capacity of temperate forest soils (e.g. Butterbach-Bahl et al., 1998; Sitaula et al., 1995; Steudler et al., 1989) due to the inhibitory effect which elevated ammonium concentrations can exert on CH_4 oxidation (for a detailed discussion also of possible stimulating effects of moderate doses of N additions on CH_4 uptake see Bodelier and Laanbroek, 2004). Furthermore, our data also show that forest management measures such as clear-cutting can significantly reduce CH_4 uptake activities throughout the soil profile. This finding is in agreement with

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results from field measurements at the Höglwald sites (Butterbach-Bahl et al., unpublished) and with results on effects of land use changes on CH_4 oxidation (Priemé et al., 1997).

5. Conclusions

This study provides for the first time data on the stratification of NO and N_2O production with soil depth under aerobic and anaerobic incubation conditions for different temperate forest sites in Germany and the Netherlands. Such data are urgently needed as guidelines for the development and testing of process-oriented models, which are able to describe the biosphere atmosphere exchange of C and N trace gases on the basis of the underlying soil microbial processes. Furthermore, our results showed that laboratory studies are very useful to detect site differences in magnitude and predominance of NO or N_2O emissions, respectively. Our observation that CH_4 uptake activity was not detectable at the Speulderbos site throughout the soil profile, most likely in consequence of chronically high rates of atmospheric N deposition, is very remarkable. It indicates that measurements of CH_4 uptake activities in the uppermost mineral soil under standardized incubation conditions may have the potential to serve as a biological indicator system for N saturation.

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Table 1. Main characteristics of the different measuring sites and dates and stratification of soil sampling.

	Höglwald-Forest			Speulderbos
	Beech	Spruce	Clearcut	Douglas fir
Location		11°10'E 48°30'N		5°41'E 52°15'N
Height above sea level (m)		540		50
Precipitation (mm yr^{-1})[a]		812[b]		768[c]
Mean annual temperature (°C)[a]		9.1[b]		10.4[c]
N-deposition via throughfall (kg N ha^{-1} yr^{-1})	~20	~30	~15	~50
Stand type	Beech	Spruce	Beech	Douglas fir
Stand age (yr)	110	100	3	
Soil type	Hapludalf	Hapludalf	Hapludalf	Hapludalf
Soil texture	Loam	Loam	Loam	Loamy sand
Sand (%)	65	64	64	62.8
Silt (%)	26	30	30	34.2
Clay (%)	9	6	6	3.0
Bulk density 0–5 cm (g cm^{-3})	0.94	1.09	1.09	0.98
C/N ratio	21.8	24.0	24.0	22.6
Organic C content (%)	5.1	2.9	2.9	3.05
Sampling dates	01/2004	11/2000, 04/2001, 08/2002, 01/2004	11/2000, 04/2001, 08/2002, 01/2004	10/2003
Sampling depths (m)				
Forest floor	0.00–0.02	0.00–0.03 0.03–0.05	0.00–0.03 0.03–0.05	0.00–0.03 0.00–0.03
Mineral soil	0.00–0.03 0.03–0.10 0.10–0.20 0.20–0.30	0.00–0.03 0.03–0.10 0.10–0.20 0.20–0.30 0.30–0.40	0.00–0.03 0.03–0.10 0.10–0.20 0.20–0.30 0.30–0.40	0.02–0.04 0.04–0.09 0.09–0.14 0.14–0.24 0.24–0.44 0.44–0.64 0.64–1.04

All soil parameters are given for 0-10 cm soil depth.

[a] Average values for the years 2002 and 2003.

[b] Given are meteorological data of the climate station Lelystad, Netherlands, which is approx. 10 km from Speulderbos. Data were provided by the Koninklijk Nederlands Meteorologisch Instituut, de Bilt, Netherlands.

[c] Given are meteorological data of the climate station Augsburg, which is approx. 20 km from Höglwald. Data were provided by the Deutscher Wetterdienst (DWD).

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Table 2. Specific rates of trace gas production across the soil profiles for the different sites (±SE). Mean trace gas production rates were divided by values for microbial biomass C. Different lower case letters indicate significant differences ($P<0.05$) between sites for a given trace gas and a given incubation condition. Different capital letters indicate significant differences ($P<0.05$) between N trace gas production for a given site (e.g. Höglwald beech site) and given incubation conditions. Different numbers indicate significant differences in N-trace gas production ($P<0.05$) between aerobic and anaerobic incubation conditions for a given N trace gas and a given site.

Trace Gas	Höglwald-Forest			Speulderbos
	Beech	Spruce	Clearcut	Douglas fir
	ng N or C mg^{-1} microbial biomass C h^{-1}			
N_2O				
Aerobic	3.51 ± 2.93^{aA1}	1.70 ± 0.53^{aA1}	0.91 ± 0.24^{aA1}	0.39 ± 0.20^{aA1}
Anaerobic	6.13 ± 2.27^{aA1}	693.9 ± 274.5^{bB2}	434.1 ± 162.5^{bcAB2}	169.4 ± 49.3^{bcB2}
NO				
Aerobic	3.31 ± 0.56^{aA1}	130.6 ± 57.7^{bB1}	31.1 ± 6.9^{aB1}	32.4 ± 16.8^{aA1}
Anaerobic	24.8 ± 6.0^{aA2}	845.8 ± 221.7^{bB2}	319.0 ± 47.5^{aAB2}	254.0 ± 87.0^{aB2}
CH_4				
Aerobic	-0.46 ± 0.34^{ab1}	-1.56 ± 0.88^{b1}	-0.50 ± 0.19^{ab1}	0.02 ± 0.06^{a1}
Anaerobic	0.07 ± 0.03^{a2}	1.14 ± 0.31^{b2}	0.69 ± 0.16^{b2}	0.12 ± 0.06^{a1}

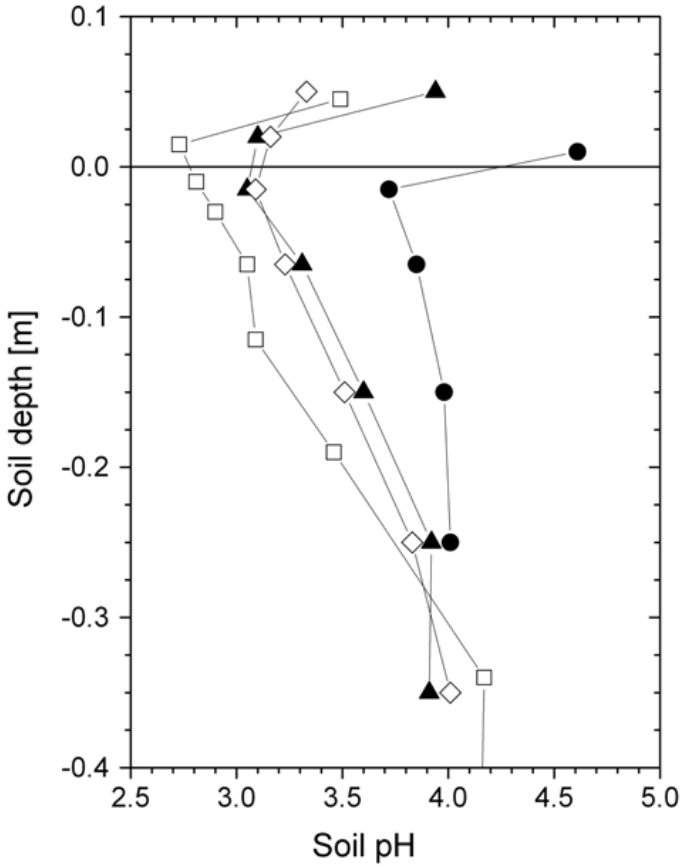

Fig. 1. Vertical distribution of soil ph values at the different sampling sites.

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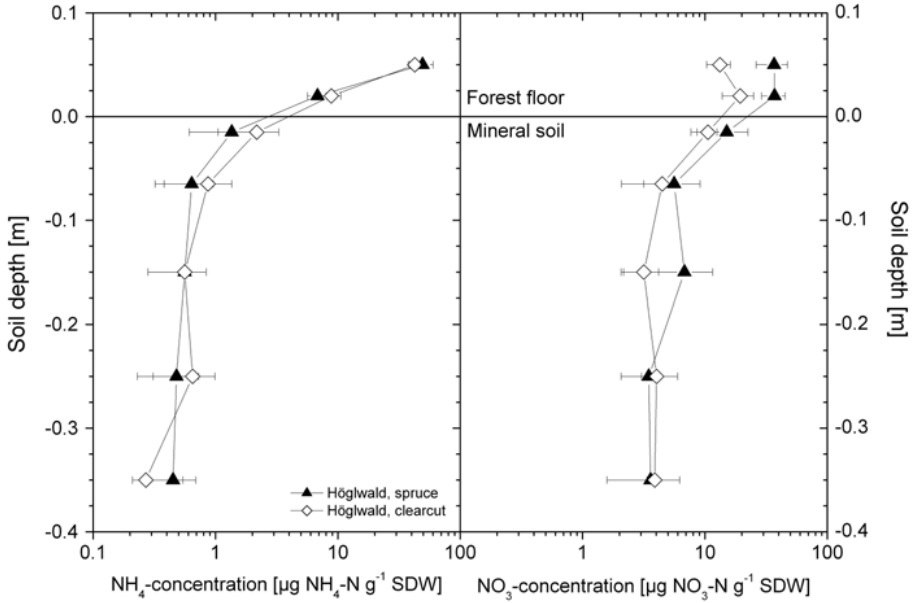

Fig. 2. Soil NO_3^- and NH_4^+ concentrations in different soil depths for the spruce and clearcut sites at the Höglwald Forest.

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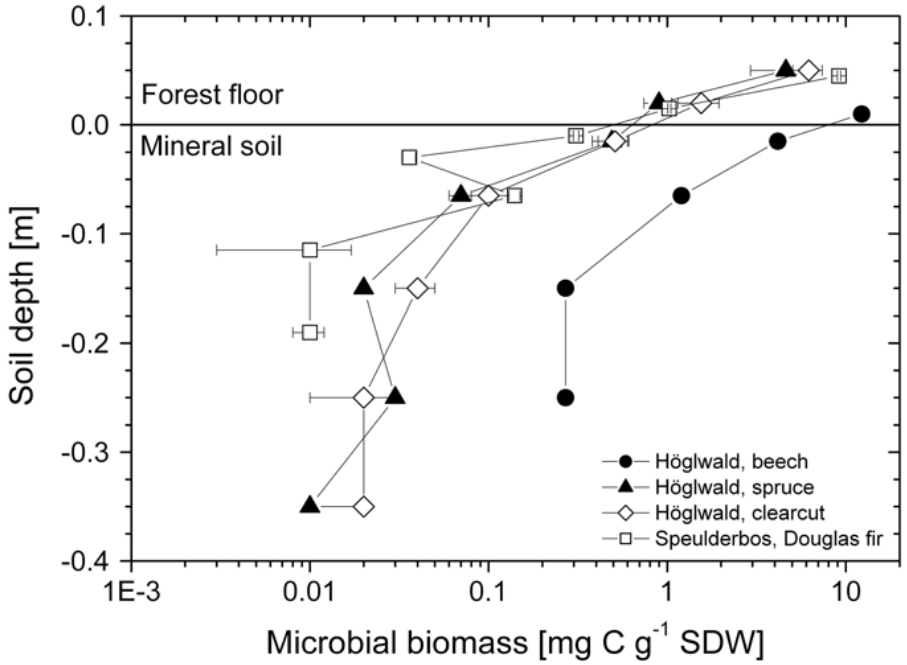

Fig. 3. Amount of microbial biomass in different soil depths at the investigated sites.

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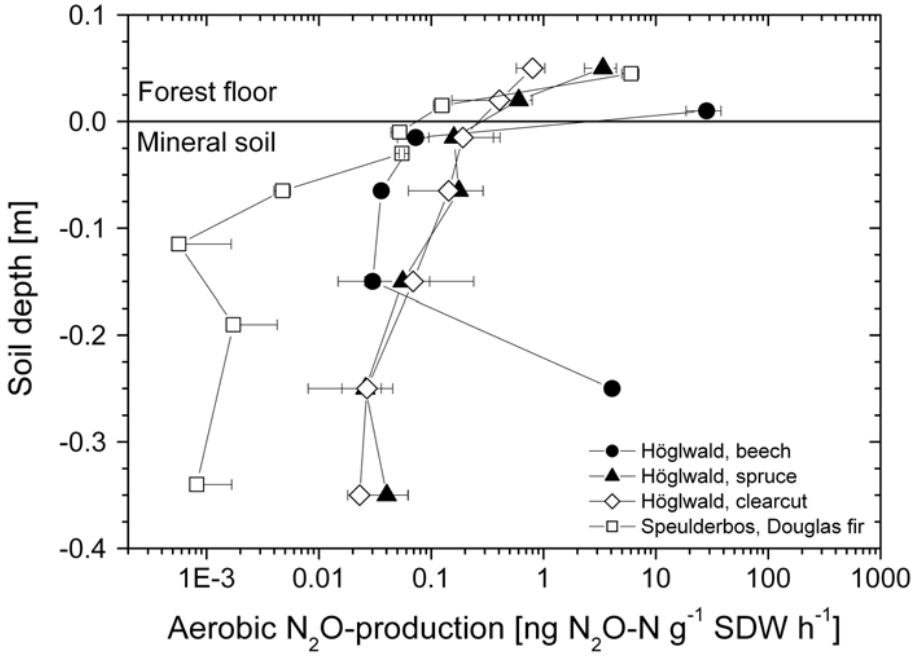

Fig. 4. N_2O production in different soil depths and at different sites under aerobic incubation conditions. Given are mean values ±SE.

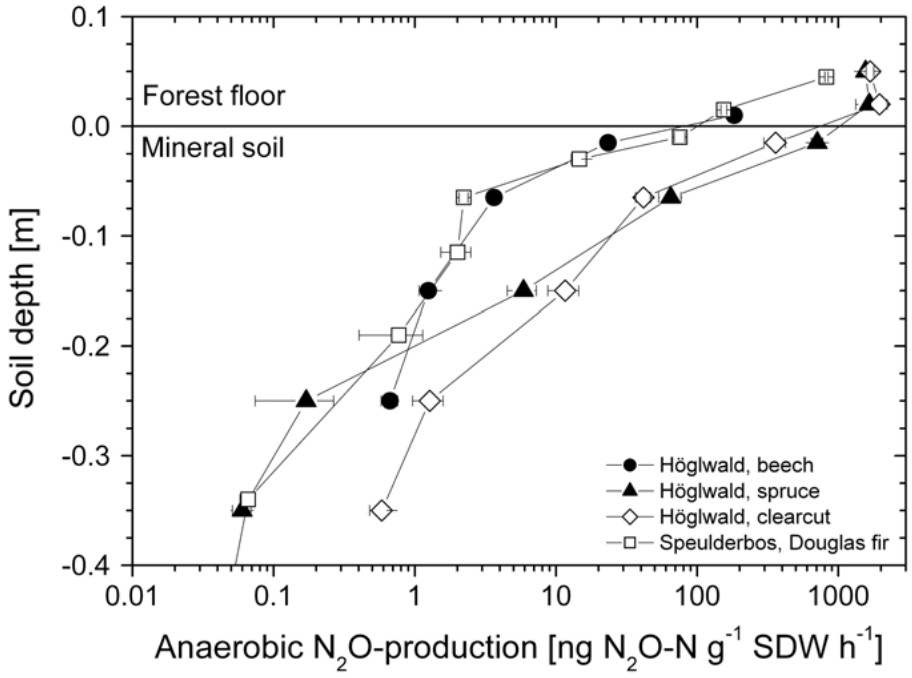

Fig. 5. N_2O production in different soil depths and at different sites under anaerobic incubation conditions. Given are mean values ±SE.

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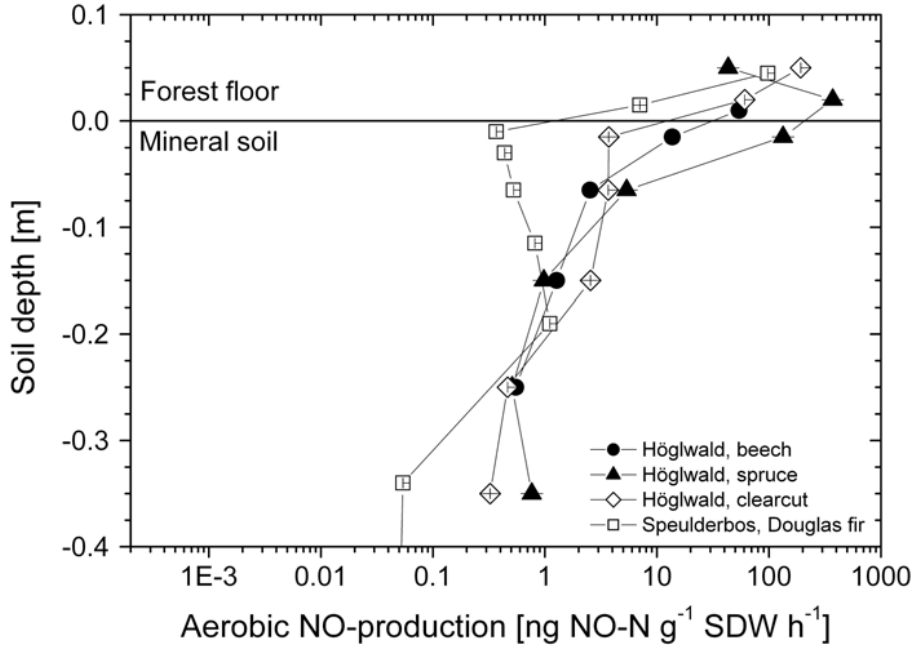

Fig. 6. NO production in different soil depths and at different sites under aerobic incubation conditions. Given are mean values ±SE.

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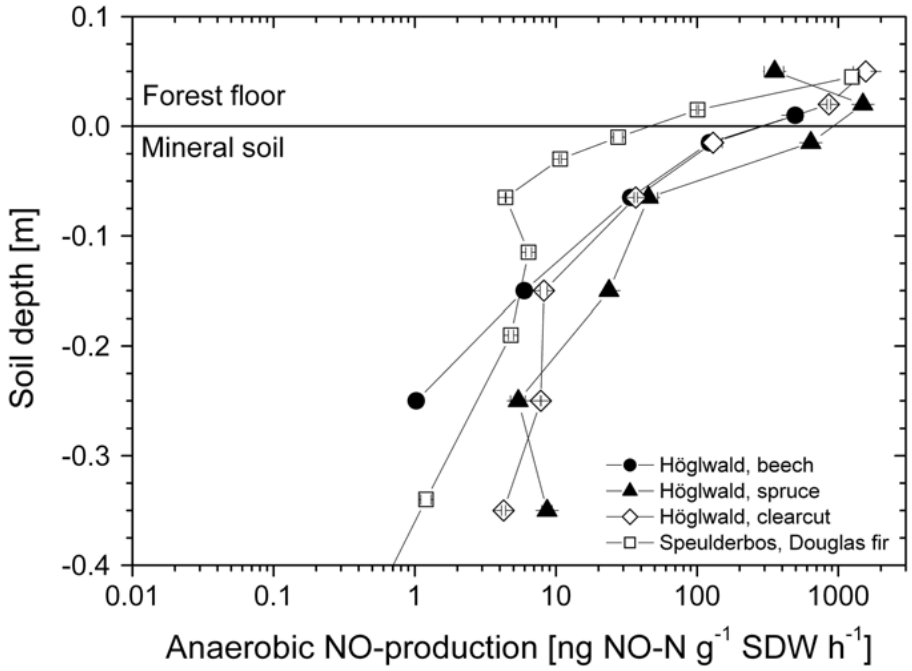

Fig. 7. NO production in different soil depths and at different sites under anaerobic incubation conditions. Given are mean values ±SE.

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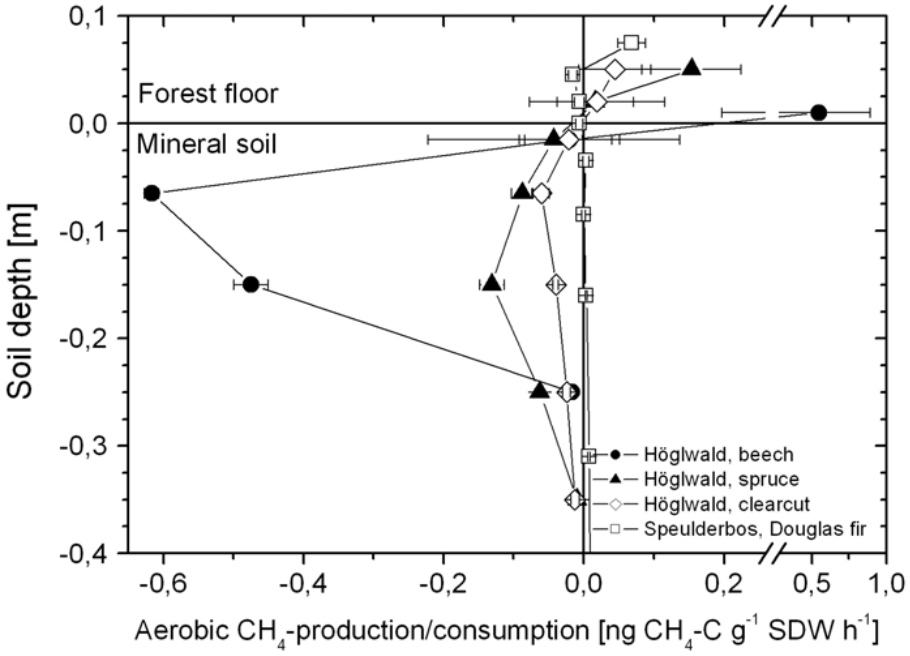

Fig. 8. CH_4 production or consumption in different soil depths and at different sites under aerobic incubation conditions. Given are mean values ±SE.

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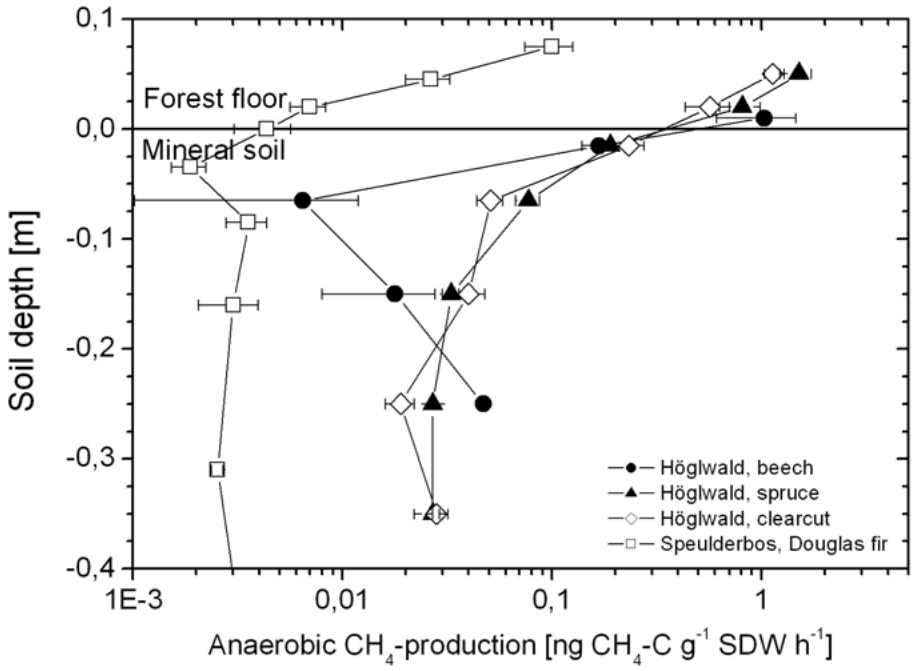

Fig. 9. CH_4 production in different soil depths and at different sites under anaerobic incubation conditions. Given are mean values ±SE.

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